Applications of IBA methods for biomineral research. Fate of metal ions released from dental works during oral use

Eugen A. Preoteasa[1], Elena S. Preoteasa[2], Santiago Gomez[3], Adela C. Scafes[1], Radu Andrei[1], Paul Ionescu[1]

[1] Horia Hulubei National Institute for Physics and Nuclear Engineering, Bucharest, Romania

[2] Helident Ltd., Bucharest, Romania

[3] Faculty of Medicine, University of Cadiz, Spain

Introduction. One major direction of our current research was focused on dental calculus or tartar, a form of hardened dental plaque caused by precipitation of minerals from saliva and gingival fluid on the teeth, which compromises the health of the gingiva. The results confirmed the assumption that certain forms of dental calculus may accumulate metal ions released by corrosion from metallic dental works. Moreover, preliminary data showed that dental polymers like acrylate and dental cements can behave also as accumulators of trace elements from the oral environment. We hereby propose the continuation of the current study of dental calculi as well as the approach of new topics on artificial dental materials (alloys, cements, restorative materials) and on their alterations during oral use, with the aim of better understanding the metal ions' fate from corrosion to accumulation. IBA methods and especially PIXE are well suited for this purpose, as supported by our recent and previous studies in the field of biomineral research [1, 2, 3].

Working hypotheses. Dental materials that can release metal ions include dental alloys from dental works (bridges, crowns, dowels, implants) and amalgams from fillings, dental cements and dental nanocomposites. Alloys are made by Si, Al, Ti, Cr, Mn, Fe, Ni, Cu, Mo, Pd, Au, etc., and amalgams mainly by Ag and Hg. Cements contain Ca and Zn, while composites and other restorative materials incorporate Si, Al, K, Ca, Sr, Zr, Ba, La, Yb, W, Bi, etc. Note that some of these metals are toxic above critical concentrations. Practically all these metals can be detected by PIXE, which can see also some relevant non-metals like P, S, Cl. In addition PIGE can detect conveniently F, Na, Mg, P. It is plausible that the metal ions once released by corrosion or slow solubilization could be bound and accumulated by dental calculi, cements and restorarive materials. Potentially relevant information can be obtained by analysis of surgically extracted dental works, which are composed of several materials (a dental alloy, a gold piece, cement and acrylate. The use of the Tandetron capability of beam focusing and positioning allows the

separate analysis of each part. If metal ions from other dental works present in the oral cavity will be released, they can accumulate in these materials, in addition to the ions released from the local environment. Complementary examination of calculus, cement and acrylate with optical microscopy techniques (polarized light, histochemical colorations) may bring additional insight on the fate of metals in the biomineral structures used in the oral cavity.

Preliminary results. One interesting result of our previous studies of dental calculus showed that proper dental calculus contained P and Ca as major elements and Zn as a minor one. The former are associated with hydroxyapatite (HA) or another calcium phosphate as the major mineral constituent, while the later is present in the active center of the enzyme alkaline phosphatase (ALP) which synthesizes HA and which belongs to certain bacteria from the dental plaque. Thus proper dental calculus of biogenic origin is identified by P, Ca and Zn. However, in preliminary PIXE examinations of a biomineral deposited on dental works and apparently similar to dental calculus, we could not evidence P, Ca and Zn but other elements instead like Si, Al, K, Ca, etc.; this material was looking like some kind of 'abiogenic dental calculus'. Morever, we observed that certain proper and 'abiogenic' dental calculus specimens, as well as cement and acrylate, behave like accumulators of metals from dental works, e.g. Cr, Mn, Fe, Ni when the dowel of a crown or bridge is made from an alloy composed of these metals. This reveals corrosion of the alloy on one side as well as migration and binding of the metallic cations, probably on anionic sites of the dental materials. We also observed that various zones of the same dental calculus had different colors, and showed affinity for different metal ions. Moreover, this spatial heterogeneity manifested itself also for different luminescence yield in different zones of certain calculi when bombarded with the 3 MV protons. The last phenomenon may be due to cathodoluminiscence (electron-hole recombination in the solid) and/or to visible fluorescence, and may yield information on the biominerals' electron band structure provided that a specific upgrading of the instrument is possible. Also, when the forthcoming microbeam mapping facility will be operational, it would provide valuable information on the heterogeneous distribution of metals in the dental calculus and materials. The study of the damage of dental materials during use by processes such as corrosion and slow dissolution might also help improving the materials and extending their lifetime.

The main advantages of the 3 MV Tandetron for biological studies in general and for biomineral structures in particular are the following: 1) the visualization of sample's surface and precise positioning of the beam; 2) the windowless detectors which allow e.g. detection of light elemets by PIXE; 3) the simultaneous detection of PIXE, PIGE and RBS spectra; and 4) the automated control of experimental parameters; and 5) the forthcoming upgrading of the Tandetron for microbeam mapping, which would be of high importance for the study of metals' distribution in the structure of dental calculi, cement and acrylate.

Experimental-setup

The Measurements will be performed at the Bucharest 3 MV Tandetron ™ in an experimental setup using three detectors, corresponding to the PIXE, PIGE and RBS methods

For PIXE, a FAST SDD®, Amptek's highest performance silicon drift detector (SDD), with a detector thickness of 500 µm, 0.5 mil thick Be window, and energy resolution of 122 eV at 5.9 keV, 25 mm^2 active area collimated to 17 mm^2, was mounted inside the experimental chamber.

For PIGE, a GEM10P4-70 coaxial gamma retractable detector (energy resolution of 1.75 keV at 1.33 MeV of 60 Co, 75 cm 3 active volume, and 10% relative efficiency) will be situated outside of the experimental chamber, at about 12-15 cm from the target [3]). For RBS, two ion-implanted silicon detectors for charged particle are available, one fixed and one movable, with a 17 keV energy esolution for a 2 MeV He beam [3].

The targets will be positioned normal on the beam direction. PIXE and PIGE detectors will be placed at 45° and RBS movable detector at 165° with respect to the beam direction and the measurements will be performed in high vacuum (10^{-6} mbar). A 3 MeV proton beam will be used and PIXE , PIGE and RBS spectra will be simultaneously detected . The beam current will be in the range of 1–2 nA, to limit sample damage.[3]

The samples, fragments of dental calculi are taken from teeth of patients and from surgically extracted crowns and bridges. These fragments of tartar are fixed with a cyanacrylic adhesive on high purity graphite planchets for electron microscopy. Some extracted dental works, consisting in alloy dowel, cement and acrylate crown are washed with deionized water, dried and analyzed by IBA without further preparation after being fixed on carbon pads. Cement and acrylate fragments are also cut from the extracted dental works with a diamond burr. Teeth roots were cleaned with an ultrasonic cleaner in order to analyze the cementum surface without dental calculus and organic contaminants. For optical microscopy, samples will be embedded in polymethylmetacrilate and cut in sections 20 – 100 mm thick. Optical microscopy and histochemistry preparations will be performed by Prof. Santiago Gomez from the University of Cadiz, Spain.

PIXE spectra are recorded with and without 20 ⬜m Al filter. The spectra are normalized to the collected charge. PIXE analysis of P and Ca is done with 2 % uncertainties; for trace elements the uncertainties are of 10 – 50 %. In PIGE the uncertainties are of 10 % for Na, Al and P, 20 % for F, and 35 – 50 % for Mg. RBS provide information about Ca, C, N and O contents. In the future, of much importance would be the upgrading of the Tandetron for microbeam mapping, which would boost the amount of available information.

Certified reference materials (CRMs) are used as standards and/or for analytical quality control including pelleted hydroxyapatite (bone ash) (NIST SRM-1400), fluorspar (NIST SRM-180), glass (NIST-611), soil (SS-P, Kosice, SK), and hay (IAEA-V10). In addition, pellets of high purity chemical compounds (KCl, NaCl, S, CaSO$_4$, CaCO$_3$, CaF$_2$, LiF, and MgO) were used for PIXE calibration and/or quantitative standardization for PIGE.

Conclusion. One can expect that PIXE supplemented with PIGE and RBS, and complemented with optical microscopy techniques, may bring a deeper insight on the fate of metal ions from dental materials and calculus, and on the mechanisms of corrosion and solubilization, as well as on pathways of migration, affinity and accumulation. The results may be useful for a better understanding of oral pathology and for improving the materials under investigation for extending their lifetime and reducing their toxicity.

Beam request*.* For the IBA experiments at the 3 MV Tandetron we request 7 days (21 samples, 2.5 hours/sample measured with and without Al foil, sitting in front of X-ray detector).).

References

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